RENAL CLEARANCE AND BRAIN TUMOR LOCALIZATION IN MICE OF ^{99m}Tc COMPOUNDS OF (Sn)DTPA, (IRON-ASCORBIC ACID) DTPA, AND IRON-ASCORBIC ACID

Tad Konikowski, Howard J. Glenn, and Thomas P. Haynie

M. D. Anderson Hospital and Tumor Institute, and The University of Texas at Houston Graduate School of Biomedical Sciences, Houston, Texas

Technetium-99m-iron ascorbic acid was first introduced as a kidney scanning agent (*1*) and then recommended for brain scanning (*2*). Later the addition of diethylenetriaminepentaacetic acid (DTPA) was thought to give improved biologic properties, and a commercial product containing iron, ascorbic acid, and DTPA for labeling with ^{99m}Tc was introduced (*3*). Another method of labeling DTPA with ^{99m}Tc using stannous tin as the reducing agent (*4*) was developed and has more recently become commercially available (*5*). This paper reports on renal clearances (*6–8*) and kidney and liver time-course distributional studies of two commercial ^{99m}Tc-DTPA chelates and of ^{99m}Tc-iron ascorbic acid in mice. Also in mice, a transplantable brain tumor has been used for studies of the pharmacokinetics of these compounds in the experimental brain tumor and related tissue time-course relationships (*9–11*).

MATERIALS AND METHODS

Radiopharmaceuticals. Technetium - 99m - (Sn) DTPA was prepared in our laboratory using a commercially available kit*. This material was prepared according to directions provided by the manufacturer. Column gel chromatography of this product indicated that more than 95% of the material was in the true DTPA chelate form (*12*).

Technetium-99m-(iron ascorbic acid) DTPA was prepared in our laboratory using a commercially available kit†. Preparations of this material usually have 90–100% of the ^{99m}Tc activity in a complexed or nonpertechnetate form as determined by thin-layer chromatography. Technetium-99m-iron ascorbic acid was prepared using ingredients from the same lot of commercial kit used for the preparation of ^{99m}Tc-(iron ascorbic acid) DTPA (Renotec) except that the DTPA was eliminated from the preparation.

Renal clearance procedure. The method for the measurement of renal clearance has been described previously in detail (*6–8*). In summary, male Yale-Swiss mice averaging 20 gm in weight and without brain tumors were employed. The mouse was induced to urinate, the penis ligated, and injections were made through the tail vein. A minimum of six mice were used for each point. At the end of the experimental time period the animals were euthanized, a heart's blood sample was obtained, and the urine quantitatively collected by dissection and intact bladder removal. Blood concentration and cumulative urine excretion curves were drawn to correlate blood levels of radioactivity with urine content. Time-course studies of the radiopharmaceuticals in liver and kidney were done simultaneously.

Mouse tumor and distribution studies. The mice employed were of the Yale-Swiss strain. The transplantable brain tumor was induced originally by methylcholanthrene in 1951 and has been carried since that time by weekly transplantations. The technique of transplantation has been described previously in detail (*9,10,13*) and may be summarized as follows: mice were studied on the eighth or ninth day following transplantation when the tumor had grown to a size of approximately 65 mg. The labeled compound was injected into the tail vein, care being taken to avoid infiltration. Mice were left undisturbed with food and water present from the time of injection to the time of tissue sampling when they were quickly euthanized. Samples of blood, tumor, brain,

* DTPA Kit, Diagnostic Isotopes, Inc., Upper Saddle River, N. J. 07458.

† Renotec, E. R. Squibb and Sons, Inc., Radiopharmaceutical Dept., New Brunswick, N. J. 08903.

Received Apr. 22, 1972; revision accepted June 26, 1972.

For reprints contact: Tad Konikowski, The University of Texas, M. D. Anderson Hospital and Tumor Institute, Houston, Tex. 77025.

muscle, and skin were removed, weighed, dissolved in nitric acid, counted, and compared with a standard representative of the total dose injected.

Evaluation of results. The average value of data obtained from at least six mice was used for each data point. For each experimental point, the Student's t-test of significance was used to evaluate differences between groups of animals with an arbitrary level of significance chosen as $p < 0.05$. The renal clearances of radiopharmaceuticals were standardized to a 1.73-m^2 surface area and compared with the clearance of inulin-^{14}C-carboxyl whose clearance closely approximates that of inulin, the accepted standard for the measurement of glomerular filtration rate. This comparison permitted an estimation of the amount of tubular excretion (TE) or tubular reabsorption (TR) associated with each compound.

RESULTS

Renal clearance. Figure 1 gives the blood disappearance curves for the three compounds in mice after single intravenous injection. Percent dose per milliliter blood is plotted against time. Individual midperiod blood values, B, are calculated from the equation

$$\frac{B_1 - B_2}{2.3 \log B_1/B_2}$$

and, as described in previous publications, the plasma value, P, is calculated by the equation

$$P = \frac{B}{1 + (RBC/P)} \times 1.805.$$

Figure 2 shows the cumulative urinary excretion curves in mice of the three labeled compounds after single intravenous injections. Since these curves represent total or cumulative urinary excretion at any time period, the urinary concentration is equal to UV in the classical clearance equation, and it is determined from the equation

$$UV = \frac{U_2 - U_1}{\text{period (min)}}.$$

Using data derived from Figs. 1 and 2, renal clearance values for blood are determined using UV/B and for plasma using UV/P. The average blood and plasma kidney clearances in mice for the three ^{99m}Tc-labeled compounds are given in Table 1. To allow for necessary "mixing time" within the mouse, clearance data from 0 to 5 min was not taken for rapidly excreted substances and from 0 to 10 min for more slowly excreted substances (7). Technetium-99m-(Sn) DTPA is cleared considerably faster than the other two compounds. For ready comparisons of the renal clearances of the three compounds,

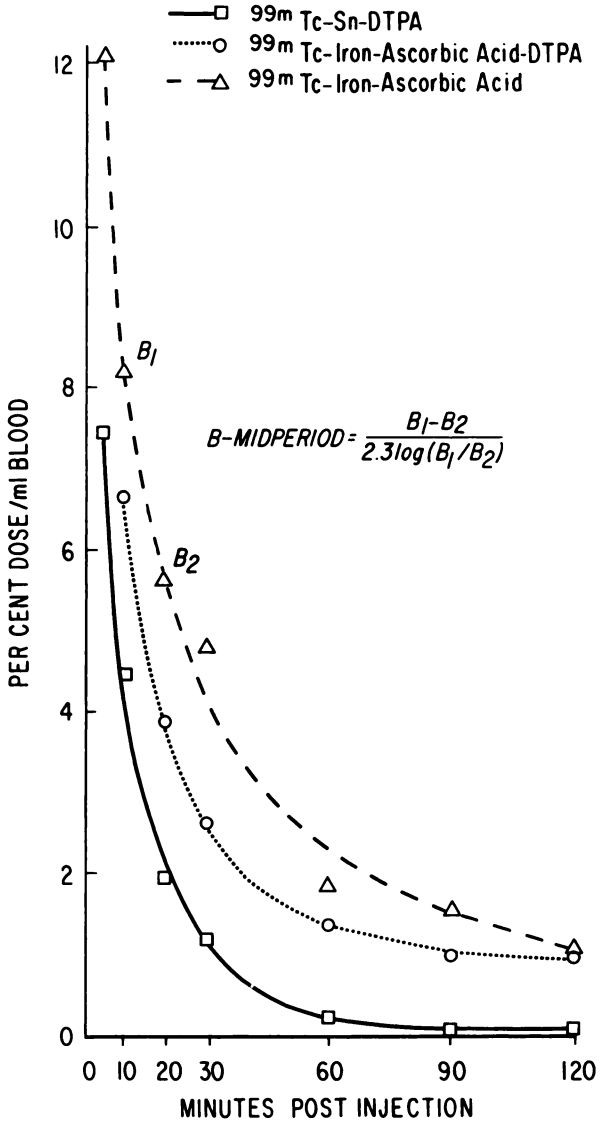


FIG. 1. Blood concentration in mice of ^{99m}Tc-labeled compounds after single intravenous injection.

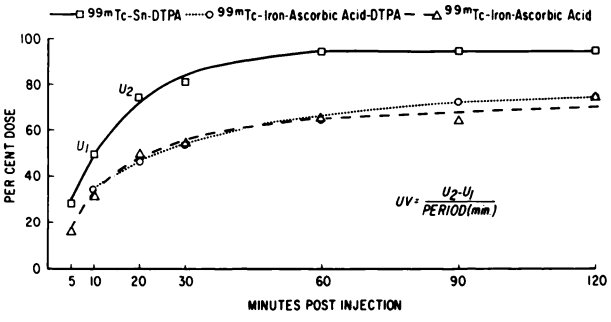


FIG. 2. Cumulative urinary excretion from mice of ^{99m}Tc-labeled compounds after single intravenous injection.

a clearance ratio graph is constructed (Fig. 3). The ratio of the average clearance of the substance at high and low concentrations to inulin is plotted against percent dose per milliliter in the plasma.

TABLE 1. AVERAGE KIDNEY CLEARANCE IN MICE OF ^{99m}Tc COMPOUNDS

	^{99m}Tc-(Sn) DTPA	^{99m}Tc-(iron ascorbic acid) DTPA	^{99m}Tc-iron ascorbic acid
Blood clearance (ml/min)	0.8331	0.2256	0.1501
Plasma clearance (ml/min)	0.5097	0.1434	0.0916

Inulin clearance (ratio 1.0) is used as a standard. Substances with an average clearance value appearing above the inulin line are cleared by glomerular filtration (GF) and TE. Those with values which appear below the inulin standard line are assumed to be reabsorbed by the tubules (TR). Sample calculations for TE and TR are given on the graph. Technetium-99m-(Sn)DTPA is cleared by GF plus TE; technetium-99m-(iron ascorbic acid) DTPA and ^{99m}Tc-iron ascorbic acid are cleared by GF followed by TR.

Table 2 lists the renal clearance evaluation indices for the three ^{99m}Tc-labeled complexes and for inulin-^{14}C-carboxyl and iodohippurate sodium ^{131}I(OIH). Clearance indices for the latter two standards have been reported in detail previously (*6,7*). All clearance values are standardized to a 1.73-m^2 surface area.

Only ^{99m}Tc-(Sn) DTPA is cleared rapidly enough to give clearance values at both high and low blood and plasma concentrations. The average plasma clearance values at low and high plasma concentrations are 138.9 and 85.6 ml/min, respectively. It is cleared 2.48 times as rapidly as inulin at low plasma concentration and 1.53 times as rapidly as inulin at high plasma concentration. The material is cleared 59.7% by TE at low plasma concentration and 34.6% by TE at high plasma concentration.

Technetium-99m-(iron ascorbic acid) DTPA is found to have an average renal plasma clearance of 29.5 ml/min at high plasma concentration. It is cleared 0.53 times as fast as inulin, with about 47% retained by tubular reabsorption.

Technetium-99m-iron ascorbic acid is cleared the

TABLE 2. ^{99m}Tc

	Labeled compound or carrier*		Average plasma concentration and range/ml	
	Body dose (μg)	Dose (μg/gm BW)	(cpm%)	(μg%)
^{14}C-inulin*	254.0	12.7	2.27 *0.74–5.88*	*5.77* *1.88–14.94*
^{131}I-Hippuran* (OIH) purified (standard) free iodide = 0.29%	20.0	1.0	L 0.45 *0.22–1.00* H 2.96 *1.80–4.12*	0.09 *0.04–0.20* 0.59 *0.36–0.82*
^{99m}Tc-(Sn) DTPA*	142.0	7.1	L 0.58 *0.35–0.86* H 4.01 *1.15–9.73*	0.82 *0.50–1.22* 5.69 *1.63–13.82*
(a)* ^{99m}Tc-iron-ascorbic acid	(a) 286.0	14.3	H 4.32 *2.12–9.07*	(a) 12.36 *6.06–25.94*
(b)* ^{99m}Tc-iron DTPA	(b) 142.0	7.1		(b) 6.13 *3.01–12.88*
(a)* ^{99m}Tc-iron	(a) 286.0	14.3	H 6.96 *3.88–23.36*	(a) 19.91 *11.10–35.35*
(b)* ^{99m}Tc-ascorbic acid	(b) 286.0	14.3		(b) 19.91 *11.10–35.35*

* The substance calculated.
† Extrapolated to a 1.73 m^2 surface area UV/B or UV/P·1.73/0.114 $W^{2/3}$ F = 206.0 for a 20-gm mouse.
L, low concentration of substance in blood or plasma/ml : : 0.1–1.0% dose.
H, high concentration of substance in blood or plasma/ml : : 1.0 and over % dose.
RBC/P, red blood cells to plasma ratio.
C_x, renal clearance of substance under investigation.

slowest of the three compounds, with a high-concentration renal plasma clearance of 18.9 ml/min. It is cleared 0.34 times as fast as inulin with an estimated 66% being reabsorbed by the tubules.

Kidney-liver studies. Figure 4 shows a time-course plot of the three compounds in the kidney of normal mice after single intravenous injections in which percent dose per total organ is plotted against time. There are significant differences between the (iron ascorbic acid) DTPA and the (Sn) DTPA compounds ($p < 0.001$) for all time periods of 20 min and over. There are significant differences ($p < 0.01$ and $p < 0.02$, respectively) between the (iron) ascorbic acid) DTPA and the iron ascorbic acid compound only at the 20- and 30-min points.

Figure 5 shows the variation of percent dose per organ (liver) with time. There are obvious significant differences between the values for the ^{99m}Tc-(Sn) DTPA and the other two ^{99m}Tc-labeled compounds. When the values for ^{99m}Tc-(iron ascorbic acid) DTPA and ^{99m}Tc-iron ascorbic acid are compared, there are significant differences ($p < 0.01$) only at the 10-, 20-, and 30-min points.

Figure 6 shows the kidney-to-liver and kidney-to-blood ratios of the three labeled compounds plotted against time. Again, the great similarity of the behavior of ^{99m}Tc-(iron ascorbic acid) DTPA and ^{99m}Tc-iron ascorbic acid can be seen. The kidney-to-liver ratios of ^{99m}Tc-(Sn) DTPA are consistently greater than the ratios of the other two compounds. The kidney-to-blood ratios of the (Sn) DTPA compound are also greater than those of the other two compounds after the 30-min period. Unfortunately, the total activity in the kidney of the (Sn) DTPA chelate is also much less.

Tumor-tissue localization of radioactivity. In Table 3 the percent of injected dose per gram with standard deviation for each of the three compounds in tumor, brain, blood, skin, and muscle at 10, 20, 30, 60, 120, 180, and 240 min after injection is seen. It is apparent that the distribution of ^{99m}Tc-(Sn) DTPA differs from that of ^{99m}Tc-(iron ascorbic acid) DTPA and ^{99m}Tc-iron ascorbic acid. This difference can be seen in Fig. 7 which shows the variation of mean percent dose per gram of tumor content with time. There are significant differences

EVALUATION INDICES

RBC/P		UV/B† clearance (ml/min)		UV/P† clearance (ml/min)		Ratio (UV/P) $\frac{C_X}{C_{IN}}$		TR(—) TE(+) (%)		UV/P deviation from OIH standard (%)	
(5 min)	(60 min)	L	H	L	H	L	H	L	H	L	H
0.00	0.00	101.2		56.1		1.00		0.00		—	
0.44	0.63	221.1	190.0 Avg. 215.4	190.6	153.3	3.36	2.73	+70.2	+63.4	Standard	
0.04	0.19	199.9	137.7 Avg. 171.6	138.9	85.6	2.48	1.53	+59.7	+34.6	—27.1	—44.2
0.10	0.20	—	46.7	—	29.5	—	0.53	—	—47.0	—	—80.8
0.06	0.17	—	31.0	—	18.9	—	0.34	—	—66.0	—	—87.7

C_{IN}, renal clearance of inulin (GFR).
TR, (tubular reabsorption) ratios of C_{IN}-$C_X \times 100$.
TE, (tubular excretion) ratios of C_X-$C_{IN}/C_X \times 100$.
The values of the labeled compounds in the plasma can be calculated from the cpm % multiplied by the body dose injected. For example, if the cpm % dose/ml plasma was 2.27 and the body dose was 254.0 μg, the average plasma concentration would be $0.0227 \times 254.0 = 5.77$ μg%.

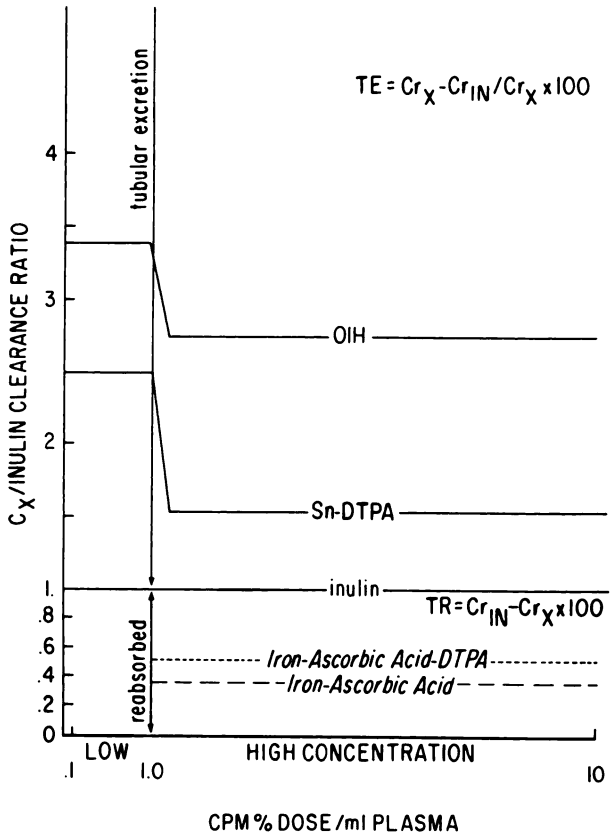


FIG. 3. Clearance ratios of ^{99m}Tc-labeled compounds compared with inulin and orthoiodohippurate.

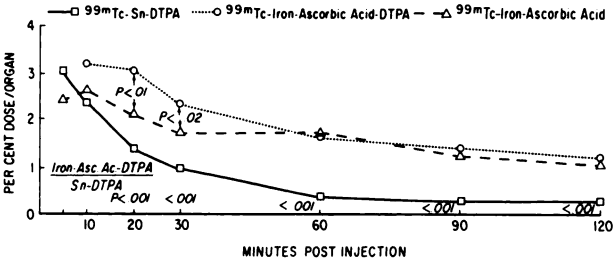


FIG. 4. Time-course plot of percent dose per organ (kidney) of ^{99m}Tc-labeled compounds.

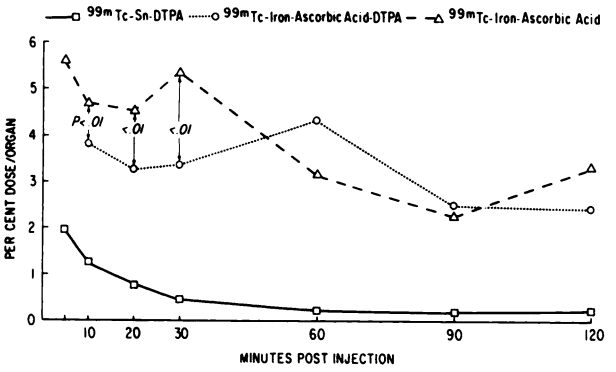


FIG. 5. Time-course plot of percent dose per organ (liver) of ^{99m}Tc-labeled compounds.

for all time periods ($p < 0.02 - p < 0.001$) between ^{99m}Tc-iron ascorbic acid and ^{99m}Tc-(Sn) DTPA, while there is significant difference ($p < 0.05$) between ^{99m}Tc-iron ascorbic acid and ^{99m}Tc-(iron ascorbic acid) DTPA only at the hour time period. The mean percent dose per gram of tumor of ^{99m}Tc-(Sn) DTPA falls below 1% beyond the 30-min point. The tumor content of the other two compounds is significantly higher for the duration of the study.

In Fig. 8 the tumor-to-tissue ratios of the three ^{99m}Tc-labeled compounds are compared. There are significant differences ($p < 0.02 - p < 0.001$) between tumor-to-brain ratio of ^{99m}Tc-iron ascorbic acid and ^{99m}Tc-(Sn) DTPA at and beyond the 30-min time period. There are apparent significant differences between ^{99m}Tc-iron ascorbic acid and ^{99m}Tc-(iron ascorbic acid) DTPA only at the 2- and 3-hr time periods.

Although there are some apparent significant differences (Fig. 8) between ^{99m}Tc-(Sn) DTPA and the other two compounds in tumor-to-blood, tumor-to-skin, and tumor-to-muscle ratios, these are of little importance because of the low tumor content of the ^{99m}Tc-(Sn)DTPA.

DISCUSSION

It is well known that chemical and biologic activities of ions are modified when used with chelating agents such as DTPA. Recently, complexes of ^{113m}In, ^{169}Yb, and ^{99m}Tc chelated with DTPA have been introduced as new radiopharmaceuticals for several diagnostic functions (*14–19*). These chelates have expanded the areas of interest developed by other investigators using labeled iron ascorbic acid complex (*1,2,20,21*). Because of the ideal radiation properties of ^{99m}Tc, there has been special interest in the chelates and complexes of this radionuclide. All three of the ^{99m}Tc complexes discussed here have been reported on for kidney scanning, renal function testing, and brain tumor imaging.

Renal studies. Harper, et al (*1*) introduced ^{99m}Tc-iron ascorbic acid complex as a renal scanning agent and showed through the use of a double-label technique that radionuclides ^{99m}Tc and ^{59}Fe remained associated and that the complex had physical and biologic characteristics different from either pertechnetate or ferric chloride. Winston, et al (*20*) made a critical evaluation of the iron ascorbic acid complex and concluded that it was an adequate but not outstanding renal scanning agent. In eight patients studied, they reported that the complex was cleared considerably slower than inulin and that the complex to inulin clearance ratio was 0.36. The average plasma renal clearance of inulin in their subjects

was 59 ml/min, and the average renal plasma clearance of the complex was 24 ml/min. An average of 19.7% of the activity in the blood was bound to the red blood cell. These figures agree surprisingly well with our mice data when the clearances are normalized to 1.73 m^2 surface area. From Table 2 it can be seen that our complex-to-inulin ratio is 0.34 based on values of inulin renal plasma clearance of 56.1 ml/min and complex renal plasma clearance of 18.9 ml/min. The red blood cell binding in our mice is 6–17% of the activity in the blood.

Hauser, et al (*21*) have reported on the renal uptake of ^{99m}Tc-iron ascorbic acid complex in man. Our work in mice (Fig. 4) shows a greater and more prolonged uptake in the kidney of the ^{99m}Tc-iron ascorbic acid and the ^{99m}Tc-(iron ascorbic acid) DTPA compounds than of the ^{99m}Tc-(Sn) DTPA.

Although our clearance values were found to be closely reproducible, they should not be compared with values in man or other species. The use of these radiopharmaceutical indices (RPF, RBF, TE, and TR) at low and high plasma concentrations is based on a simplified view of the complex clearance function of the kidney. This view does not take into consideration such complicating factors as plasma protein binding, bidirectional tubular transport, simultaneous secretion and reabsorption, and others. These complicating factors may be necessary in detailed clearance studies but are not necessary in the use of clearance as a bioassay for evaluating and comparing radiopharmaceuticals. Klopper, et al (*18*) have worked with dogs and humans in the evaluation of ^{99m}Tc-(Sn) DTPA, and concluded that it can be used for estimating glomerular filtration rate. Our radiopharmaceutical indices show that in mice 59.7% of the ^{99m}Tc-(Sn) DTPA is cleared by tubular excretion.

In a series of papers (*22–24*), the Brookhaven group has made other intercomparisons of this group of labeled compounds. Using Sephadex column chromatography, it has been shown that ^{99m}Tc-(Sn) DTPA is a true chelate with over 95% of the activity chelated by the DTPA. In their studies, ^{99m}Tc-(iron ascorbic acid) DTPA had only 12–22% of the activity chelated by DTPA and 9–16% in the form of pertechnetate. Our clearance data in mice can be used to estimate the amount of chelation in ^{99m}Tc-(iron ascorbic acid) DTPA. If one assumes the mouse renal plasma clearance of ^{99m}Tc-(Sn) DTPA (85.6 ml/min) to be the clearance of a completely chelated substance (*12*), and the renal plasma clearance of ^{99m}Tc-iron ascorbic acid complex (18.9 ml/min) to be the baseline clearance of iron ascorbic acid

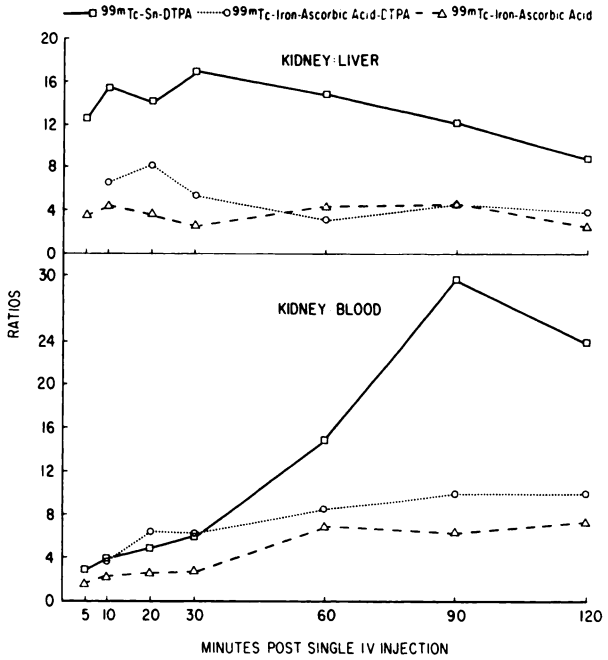


FIG. 6. Kidney-to-liver and kidney-to-blood ratios (percent dose per gram per percent dose per gram) with time of ^{99m}Tc-labeled compounds.

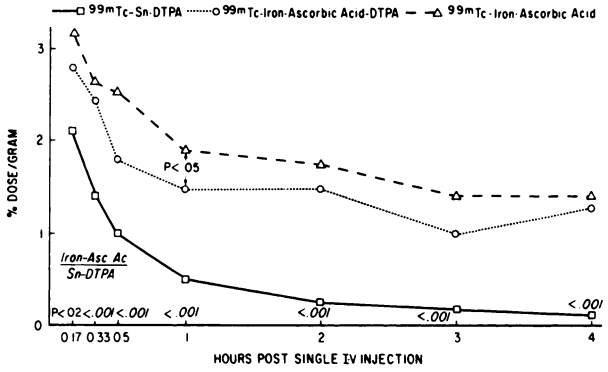


FIG. 7. Time-course plots of percent dose per gram of tumor of ^{99m}Tc- labeled compounds.

complex, then the difference in plasma clearance between the latter and ^{99m}Tc-(iron ascorbic acid) DTPA should be the contribution of the DTPA chelate. Setting up a simple proportion, it can be estimated, based on clearance data, that an average of 12.4% of ^{99m}Tc-(iron ascorbic acid) DTPA is in the form of true DTPA chelate.

Brain tumor studies. Atkins, et al (*22*) have shown that ^{99m}Tc-(Sn) DTPA is essentially identical to a ^{99m}Tc-(Fe) DTPA prepared earlier (*16*) in physical and biologic properties. Of the former, they state that their experience with the compound had been so favorable that it is now used routinely as a brain scanning agent. The experience of Brookeman, et al (*17*) and of Haynie, et al (*25*) with ^{99m}Tc-(iron as-

TABLE 3. COMPARISON OF TECHNETIUM IMPLANTED SARCOMA IN PERCENT

	Post-single-intravenous injection (hr)								
	0.17			0.33			0.5		
	I	II	III	I	II	III	I	II	III
Tumor	2.09 ±0.26	2.79 ±0.54	3.19 ±0.85	1.41 ±0.31	2.44 ±0.77	2.63 ±0.48	0.98 ±0.39	1.80 ±0.81	2.52 ±0.55
Brain	0.22 ±0.05	0.39 ±0.08	0.38 ±0.10	0.21 ±0.12	0.25 ±0.07	0.28 ±0.04	0.26 ±0.08	0.21 ±0.03	0.30 ±0.10
Blood	3.66 ±0.44	9.52 ±3.30	11.03 ±2.89	1.40 ±0.38	3.71 ±0.76	5.29 ±0.88	1.09 ±0.26	2.94 ±0.74	3.85 ±0.84
Skin*	4.30 ±1.16	8.23 ±2.24	5.75 ±0.77	1.84 ±0.61	3.39 ±0.80	3.89 ±0.69	1.15 ±0.53	2.77 ±0.60	2.99 ±1.48
Muscle	1.21 ±0.25	2.82 ±0.73	2.57 ±0.48	0.53 ±0.23	1.06 ±0.29	1.29 ±0.13	0.37 ±0.11	1.03 ±0.39	1.10 ±0.40

* Corrected for 30% fur. [for 100% skin multiply by 1.43 (100/70)].
(I) ^{99m}Tc-Sn DTPA; (II) ^{99m}Tc-iron-ascorbic acid DTPA; (III) ^{99m}Tc-iron ascorbic acid.
Each data point is the average of six animals with standard deviation.

corbic acid) DTPA as a brain scanning agent have also been favorable. Stapleton, et al (*2*) call the ^{99m}Tc-iron ascorbic acid complex a good brain scanning agent. In our test system, this complex shows the best and most prolonged tumor uptake with the ^{99m}Tc-(iron ascorbic acid) DTPA complex giving quite similar results. The tumor uptake of ^{99m}Tc-(Sn) DTPA is the poorest and the activity disappears from the tumor the most rapidly. The average tumor-to-brain tissue ratios of ^{99m}Tc-iron ascorbic acid and ^{99m}Tc-(iron ascorbic acid) DTPA are also similar and are better than the tumor-to-brain ratio of ^{99m}Tc-(Sn) DTPA. Based on these observations, it would appear that ^{99m}Tc-iron ascorbic acid without DTPA merits further investigation as a brain scanning agent.

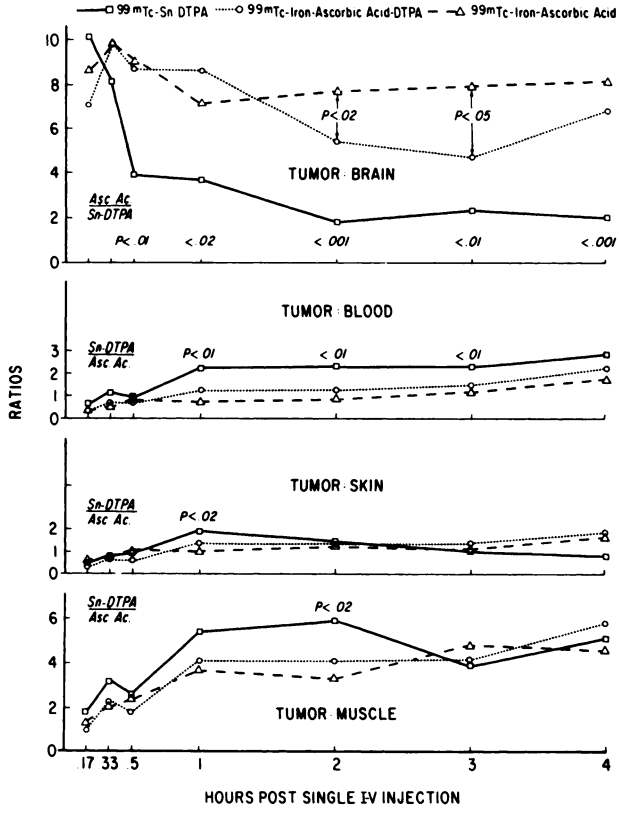


FIG. 8. Tumor-to-brain, tumor-to-blood, tumor-to-skin, and tumor-to-muscle ratios with time of ^{99m}Tc-labeled compounds.

The advantage of our study over previous investigations (*15,26*) is that we have evaluated tumor uptake in mice using implantations of tumor directly into the brain whereas most other authors have used tumors planted subcutaneously. The validity of using a subcutaneous transplantable tumor as a model for intracranial neoplasms in man for the comparison of scanning agents has been a continuing source of concern and is discussed by Soloway, et al (*27*). One of the advantages of having the tumor implanted into the brain is that the pressure and vasculature are more analogous to that of the clinical condition than when a tumor is implanted subcutaneously. Our tumor-to-brain ratios are consistently lower than those obtained with subcutaneously transplanted tumors. We also have previously emphasized the importance of the ligature technique (*9*) to prevent redistribution of blood and isotope after death. Without use of the ligature our tumor-to-brain ratios are almost double those obtained with it.

In Table 4, we have added these compounds to our list of brain scanning agents and related maximum tumor uptake in mice (irrespective of time) to average blood renal clearance (*11*). There is a continued striking inverse relationship. The slower the blood clearance, the greater the maximum tumor uptake. We have commented on this previously.

COMPOUNDS IN VARIOUS TISSUES IN MOUSE BRAIN— OF INJECTED DOSE/GRAM

Post-single-intravenous injection (hr)											
1			2			3			4		
I	II	III	I	II	III	I	II	III	I	II	III
0.47	1.48	1.89	0.25	1.49	1.75	0.17	1.02	1.40	0.11	1.29	1.41
±0.24	±0.30	±0.23	±0.06	±0.39	±0.31	±0.04	±0.31	±0.36	±0.04	±0.19	±0.45
0.14	0.18	0.28	0.15	0.31	0.23	0.08	0.22	0.19	0.07	0.24	0.19
±0.07	±0.05	±0.08	±0.07	±0.14	±0.04	±0.03	±0.07	±0.07	±0.04	±0.13	±0.07
0.23	1.25	2.72	0.12	1.28	2.30	0.08	0.69	1.26	0.04	0.66	0.88
±0.15	±0.17	±0.56	±0.06	±0.32	±0.40	±0.01	±0.09	±0.10	±0.01	±0.29	±0.16
0.26	1.12	1.92	0.18	1.29	1.46	0.19	0.72	1.42	0.15	0.75	0.90
±0.13	±0.25	±0.32	±0.06	±0.71	±0.12	±0.07	±0.13	±0.50	±0.04	±0.31	±0.22
0.10	0.40	0.55	0.05	0.38	0.54	0.05	0.24	0.29	0.02	0.25	0.31
±0.09	±0.11	±0.18	±0.02	±0.12	±0.12	±0.02	±0.05	±0.04	±0.01	±0.11	±0.04

TABLE 4. RELATION OF BRAIN TUMOR UPTAKE TO RENAL CLEARANCE IN MICE

Tumor scanning agents	Highest % dose/gm tumor	UV/B clearance* (ml/min)	Maximum tumor: brain ratios
$^{99m}TcO_4^-$ (perchlorate predose 3.0 μg/gm BW)	5.14	3.7	9.7
$^{99m}TcO_4^-$	3.93	6.2	7.3
^{197}Hg-chlormerodrin	3.67	8.2	14.5
^{197}Hg-chlormerodrin (meralluride predose 0.56 μg Hg/gm BW)	3.24	11.6	14.2
^{99m}Tc-iron ascorbic acid†	3.19	31.0	9.8
^{99m}Tc-iron-ascorbic acid DTPA†	2.79	46.7	9.9
^{113m}In-DTPA	2.18	136.6	10.0
^{99m}Tc-Sn DTPA	2.09	171.6	10.1
^{169}Yb-DTPA	2.05	275.9	9.4

* Extrapolated to 1.73 m^2 surface area.
† Commercial kit, same batch.

SUMMARY

Studies of the experimental tumor localization and renal clearance in mice of ^{99m}Tc-(Sn)DTPA, ^{99m}Tc-(iron ascorbic acid)DTPA, and ^{99m}Tc-iron ascorbic acid reveal significant differences between ^{99m}Tc-(Sn)DTPA and the other compounds. The latter show slower renal clearances and higher tumor nuclide content and tumor-to-brain ratios. There is little difference in the other tumor-to-tissue ratios. The two compounds containing iron and ascorbic acid show greater and more prolonged uptake by the kidney and liver. These findings support the contention that ^{99m}Tc-(iron ascorbic acid)DTPA is not more than a few percent true chelate and is biologically closely related to ^{99m}Tc-iron ascorbic acid. Though care must be taken in extrapolating results in mice to humans, these results also suggest that there may be some advantages of the latter two compounds over the true DTPA chelate in both kidney and brain tumor localization studies.

ACKNOWLEDGMENT

This work has been supported in part by American Cancer Society Grant No. ACS-IN-43-L.

REFERENCES

1. HARPER PV, LATHROP KA, GOTTSCHALK A, et al: Pharmacodynamics of some technetium-99m preparations. In *Radioactive Pharmaceuticals,* Andrews GA, Kniseley RM, Wagner HN, eds, USAEC Symposium Series 6, CONF-651111, Springfield, Va, National Bureau of Standards, 1966, pp 335–358

2. STAPLETON JE, ODELL RW, MCKAMEY MR: Technetium iron ascorbic acid complex: A good brain scanning agent. *Amer J Roentgen* 101: 152–156, 1967

3. *Renotec Technetium-99m-Diethylenetriaminepentaacetic Acid (DTPA) Kit.* ER Squibb and Sons, Inc, New Brunswick, N J

4. ECKELMAN W, RICHARDS P: Instant ^{99m}Tc-DTPA. *J Nucl Med* 11: 761–762, 1970

5. *DTPA Kit (Diethylenetriaminepentaacetic Acid).* Diagnostic Isotopes, Upper Saddle River, N J

6. KONIKOWSKI T, HAYNIE TP, FARR LE: Inulin clearance in mice as a standard for radiopharmaceutical bioassay. *Proc Soc Exp Biol Med* 135: 320–324, 1970

7. KONIKOWSKI T, HAYNIE TP, GLENN HJ, et al: Iodohippurate sodium ^{131}I(OIH) clearance in mice. Bioassay of radiopharmaceuticals. *Proc Soc Exp Biol Med* 137: 1343–1351, 1971

8. KONIKOWSKI T, GLENN HJ, HAYNIE TP, et al: Renal clearance in mice as a bioassay for radiopharmaceuticals: Intercomparison of commercial sources of iodohippurate sodium ^{131}I(OIH). *Int J Nucl Med Biol*: to be published

9. KONIKOWSKI T, HAYNIE TP: The effect of perchlorate

on the localization of ^{99m}Tc-pertechnetate in a mouse brain sarcoma. *J Nucl Med* 11: 443–448, 1970

10. KONIKOWSKI T, HAYNIE TP: The effect of meralluride blocking on localization of radiochlormerodrin in a mouse brain tumor (sarcoma). *Int J Appl Radiat* 21: 711–718, 1970

11. HAYNIE TP, KONIKOWSKI T, GLENN HJ: The kinetics of ^{99m}Tc-, ^{113m}In-, ^{169}Yb-DTPA compounds in brain sarcoma and kidneys of mice. *J Nucl Med* 13: 205–210, 1972

12. LIEBERMAN E: Personal communication

13. FARR LE, KONIKOWSKI T: The effect of regional thermal neutron exposure upon the growth and transplantability of a malignant tumor in the mouse. In *Biological Effects of Neutron and Proton Irradiation,* vol 2, Vienna, IAEA, 1964, pp 157–172

14. CLEMENTS JP, WAGNER HN, STERN HS, et al: Indium-113m diethylenetriaminepentaacetic acid (DTPA): A new radiopharmaceutical for brain scanning. *Amer J Roentgen* 104: 139–144, 1968

15. HOSAIN F, REBA RC, WAGNER HN: Ytterbium-196 diethylenetriaminepentaacetic acid complex: A new radiopharmaceutical for brain scanning. *Radiology* 91: 1199–1203, 1968

16. HAUSER W, ATKINS HL, NELSON KG, et al: Technetium-99m DTPA: A new radiopharmaceutical for brain and kidney scanning. *Radiology* 94: 679–684, 1970

17. BROOKEMAN VA, WILLIAMS CM: Evaluation of ^{99m}Tc-DTPA acid as a brain scanning agent. *J Nucl Med* 11: 733–738, 1970

18. KLOPPER JF, HAUSER W, ATKINS HL, et al: Evaluation of ^{99m}Tc-DTPA for the measurement of glomerular filtration rate. *J Nucl Med* 13: 107–110, 1972

19. HISADA KI, MISHIMA T, HIRAKI T, et al: Intravenous radioisotope angiography using ^{113m}In Fe DTPA-ascorbic acid. *Radiology* 91: 1204–1207, 1968

20. WINSTON MA, HALPERN SE, WEISS ER, et al: Critical evaluation of ^{99m}Tc-Fe ascorbic acid complex as a renal scanning agent. *J Nucl Med* 12: 171–175, 1971

21. HAUSER W, ATKINS HL, RICHARDS P: Renal uptake of ^{99m}Tc-iron ascorbic acid complex in man. *Radiology* 101: 637–641, 1971

22. ATKINS HL, CARDINALE KG, ECKELMAN WC, et al: Evaluation of ^{99m}Tc-DTPA prepared by three different methods. *Radiology* 98: 674–677, 1971

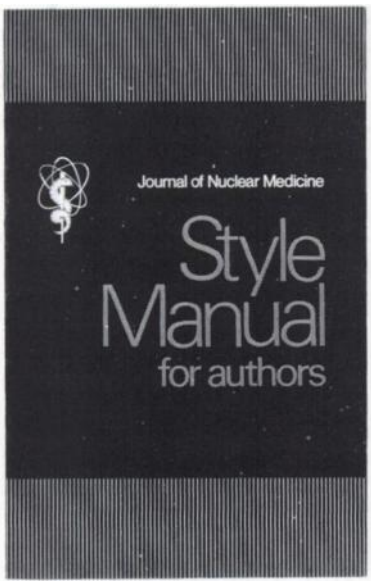
23. ECKELMAN WC, RICHARDS P, HAUSER W, et al: ^{99m}Tc-DTPA preparations. *J Nucl Med* 12: 699, 1971

24. ECKELMAN WC, RICHARDS P: Analytical pitfalls with ^{99m}Tc-labeled compounds. *J Nucl Med* 13: 202–204, 1972

25. HAYNIE TP, KONIKOWSKI T, JHINGRAN SG, et al: Brain scintigrams with ^{99m}Tc-iron DTPA complex in experimental and metastatic neoplasms: Comparison with radioactive chlormerodrin and pertechnetate. *J Nucl Med* 11: 324–325, 1970

26. SCHWARTZ ML, TATOR H: Shortcomings of ^{99m}Tc-pertechnetate as a tracer for brain tumor detection as shown by well counting of human brain tumors and a mouse ependymoblastoma. *J Nucl Med* 13: 321–327, 1972

27. SOLOWAY AH, ARONOW S, KAUFMAN C, et al: Penetration of brain and brain tumor. VI. Radioactive scanning agents. *J Nucl Med* 8: 792–799, 1967



STYLE MANUAL FOR AUTHORS NOW AVAILABLE

To help authors prepare manuscripts for publication in the *Journal of Nuclear Medicine,* the Society has just published a *Style Manual for Authors.* It includes sections on the publication process, types of manuscripts accepted, the manuscript format, style considerations, and a sample manuscript.

Make use of this handy tool! Order your copy now by sending $1.00 to:

Production Editor
Journal of Nuclear Medicine
211 East 43 Street
New York, New York 10017

Make checks payable to: The Society of Nuclear Medicine

All orders must be prepaid.